CleanAmp™ Multiplex PCR 2X Master Mix

Catalog # L-5103

L-5103-100 (100 reactions)
L-5103-BK (Bulk amount)

CleanAmp™ Multiplex PCR 2X Master Mix is an optimized, ready-to-use mix of CleanAmp™ dNTPs and *Taq* DNA polymerase in reaction buffer suited for multiplex amplification. CleanAmp™ Multiplex PCR 2X Master Mix has been utilized in assays of fifty targets and higher. Simply add primers, template DNA and water.

QC Analysis

Functional Assay; Pass
Tested in standardized PCR assay for efficiency and specificity.

Handling & Use

Store at -20°C
Stable to 15 freeze-thaw cycles. Exposure to ambient temperatures during shipping does not adversely affect product performance.

Protocols

Endpoint Muliplex PCR (25 µL)

1. Thaw CleanAmp™ Mulitplex PCR 2X Master Mix, primers and DNA template and place on ice.
   Note: Do not vortex CleanAmp™ Multiplex PCR 2X Master Mix. Mix thoroughly by pipetting up and down and collect by pulse centrifugation.
2. Prepare a reaction mixture containing all components except for the DNA template. Add CleanAmp™ Multiplex PCR 2X Master Mix, primers and sterile de-ionized water as shown in Table 1 into thin-walled PCR tubes.[1] Keep on ice.
3. Mix the reaction mixture gently to protect the enzyme, by pipetting up and down. Do not vortex. Pulse spin if necessary.
4. Add the appropriate volume of template DNA to reach a reaction volume of 25 µL.
5. Pulse spin to remove bubbles and collect reaction solution at bottom of PCR tube.
6. Place the tubes into a thermal cycler with a heated lid and perform the appropriate cycling conditions for standard thermal cycling:
   95°C for2 min
   [95°C for 15 sec; 48-60°C[2] for 30 sec; 72°C for 0.5-2 min]
   30-40 cycles
   72°C for 5 min
7. Analyze an aliquot of the completed reaction by agarose gel electrophoresis.

Table 1

Component	Final Concentration (25 µL reaction)	Volume per reaction
CleanAmp™ Mulitplex PCR 2X Master Mix	1X	12.5 µL
Forward/Reverse Primer	50-500 nM	Variable
DNA Template	Variable	Variable
Sterile De-ionized Water	Up to 25 µL	Up to 25 µL
Total Volume (µL)	**25 µL**	**25 µL**

[1] For challenging multiplex amplifications, performance can sometimes be improved by the addition of 10-30 mM KCl into the reaction.
[2] The annealing temperature should be chosen for optimal PCR performance. Most primer design software recommends an annealing temperature. The annealing temperature can also be optimized experimentally by using a thermal cycler with gradient functionality or by performing sequential experiments in which the annealing temperature is varied.

Real-Time PCR

CleanAmp™ Multiplex PCR 2X Master Mix has been successfully adapted for real-time detection using intercalating dye and probe-based detection. Please refer to the instrument manufacturer for specific protocols.

Real-time PCR may be proprietary. No license is conveyed expressly or by implication to the purchaser by purchase of any TriLink BioTechnologies products.